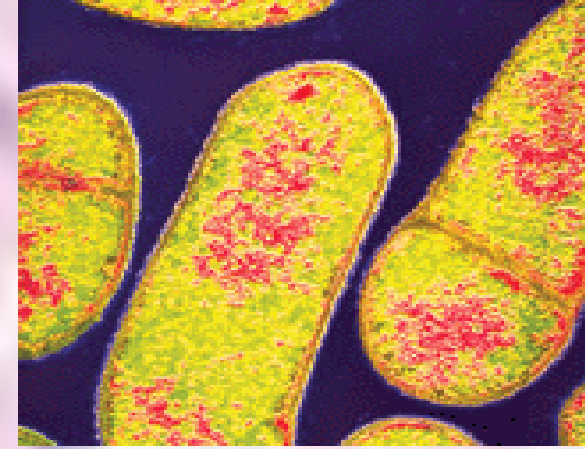




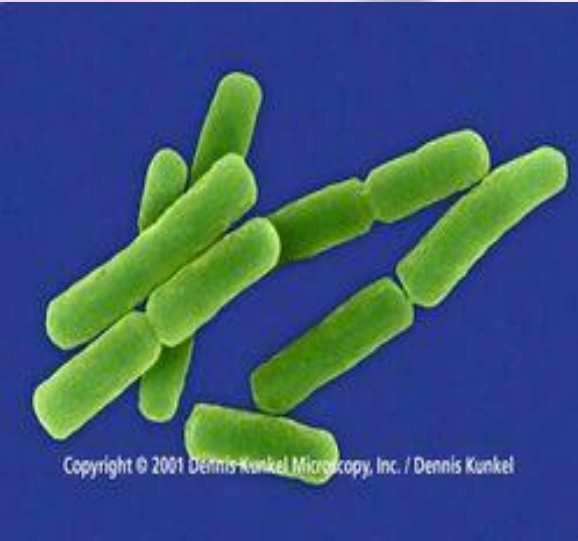
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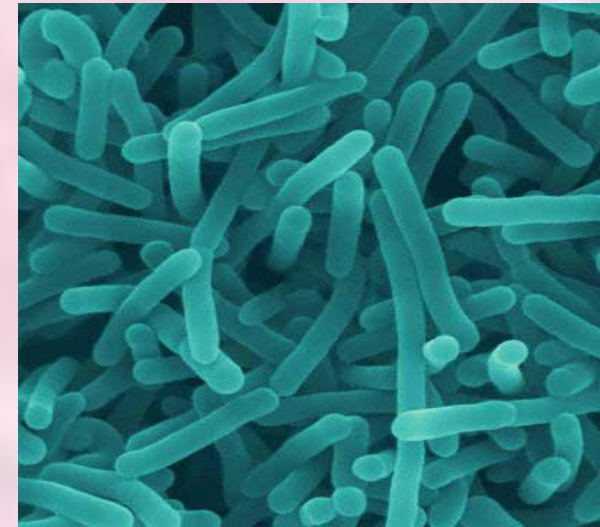
Gram Positive Bacilli

Page 48- 65

Midterm # 2 is on pages 48-90



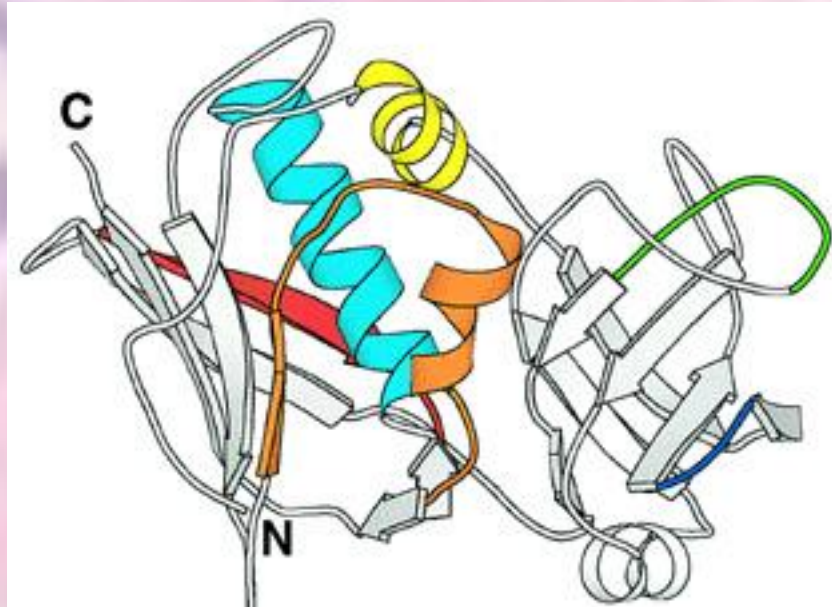
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Some bacteria are able to go into a state of hibernation (become a spore)
In the state of a spore, the bacteria is resistant to dryness, UV, disinfectant
If a bacteria cannot turn into a spore it is called a vegetative cell

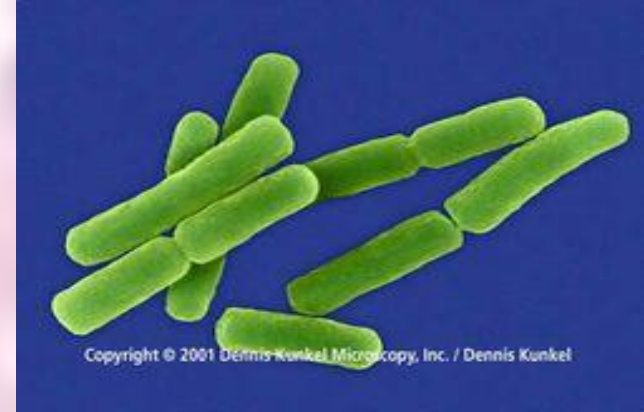
Spore Forming Rods

- *Bacillus* and *Clostridium*
- Release of potent exotoxins causes disease



Do not have endotoxins because they are gram positive and therefore do not have the extra outer layer

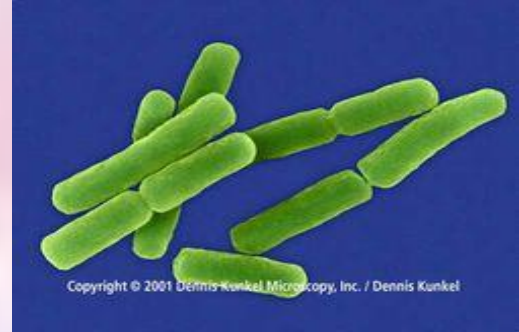
Bacillus anthracis



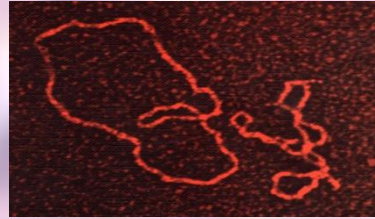
- Causative agent of anthrax
- UNIQUE protein capsule, antiphagocytic
The capsule is made out of protein (as opposed to most that are made out of sugar) *****midterm*****
- Aerobic growth conditions
- Spores are very stable, resistant to heat, drying, UV and disinfectants; spores germinate and toxins are made
- Humans exposed to spores usually through contact with animals or soil
- Used in bio-terrorism and warfare



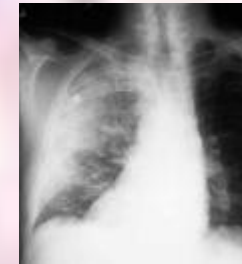
Bacillus anthracis exotoxin



- Encoded on pXO1 plasmid
encodes the 3 subunits



- Plasmid contains virulence factors which are transcribed optimally @ 37°C, increased CO₂ and serum proteins
- ?? Where are these conditions found ??
In the lungs



- Exotoxin composed of 3 separate proteins:
 - Edema factor (EF) (swelling)
 - Protective antigen (PA) Protects exotoxin from disintegration and other stuff
 - Lethal factor (LF)
 - Separately proteins are not toxic, but combined are lethal
- pXO2 plasmid encodes capsule genes
- ****BOTH plasmids required for virulence****

Get turned on in the right conditions (usually once in the lungs)

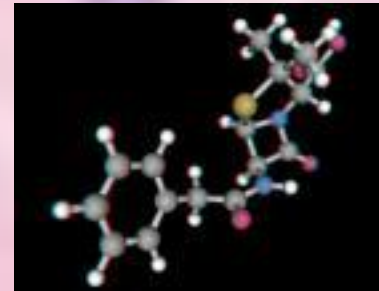
Always know: Is there a treatment? Is there a vaccine? for every illness we talk about

Prevention and Treatment

- *RAPID* treatment is essential
- Antibiotics: penicillin, doxycyclin, ciprofloxacin or levofloxacin
Do not need to remember different antibiotics
- Vaccine against PA protein is available



2 plasmids:
Remember which encodes
for what



PENICILLIN

People have figured out how to put the three components together to artificially create anthrax and to make it into a powder to send to people. (can be used for terrorist attacks)

They have figured out how to pinpoint where the anthrax has come from (like which country)

Bacillus cereus

Know the difference between the two Bacillus

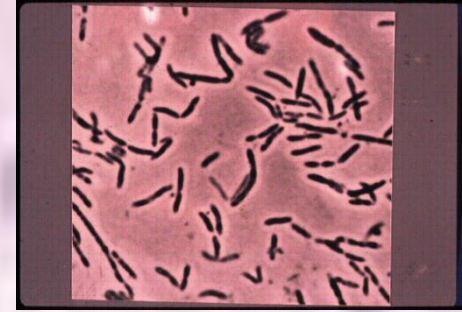


- Causes food poisoning when spores enter food product



- Motile, non-encapsulated, resistant to penicillin
- **ENTEROTOXIN** is responsible for illness Enterotoxin is an exotoxin that is made when the bug is inside your stomach
- 2 types of enterotoxin
 - (i) Heat labile: nausea, abdominal pain, diarrhea. Lasts 12-24hrs
 - (ii) Heat stable: SEVERE nausea and vomiting, short incubation
- Antibiotic treatment useless....preformed toxins

Clostridium



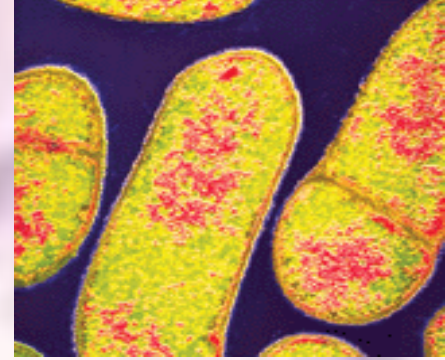
- Anaerobic → differentiates this bacteria from other spore-forming bacilli

Still spore-forming

- Botulism, tetanus, gas gangrene and pseudomembranous colitis

- POWERFUL EXOTOXINS → RAPID DIAGNOSIS

Clostridium botulinum



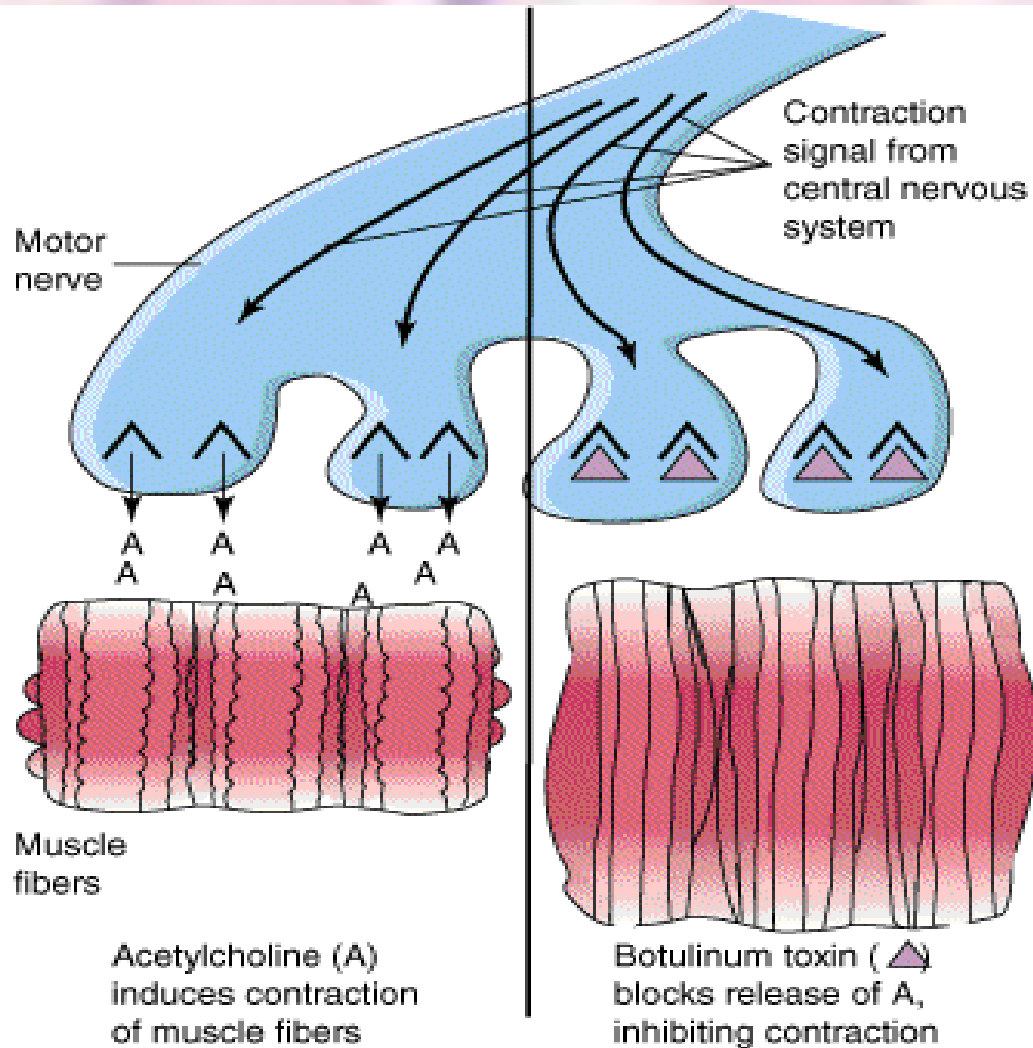
- Rapidly fatal food poisoning from lethal neurotoxin

Neurotoxin means that the toxin affects your nervous system

- Neurotoxin blocks Ach release in autonomic system; flaccid muscle paralysis:
 - Afebrile, bilateral cranial nerve palsies, double vision, trouble swallowing, muscle weakness
 - Respiratory paralysis → DEATH
 - Treatment: antitoxin and respiratory assistance
- Smoked fish, improperly canned vegetables Very rare in canned food
 - Proper cooking destroys spores



Action of Botulinum Neurotoxin



Antibodies try to pull out the neurotoxin but the neurons have a strong hold on it

Infant Botulism

- Honey contamination with spores
- Spores germinate and bacteria colonizes intestine
- Neurotoxin release



2-3 days of constipation



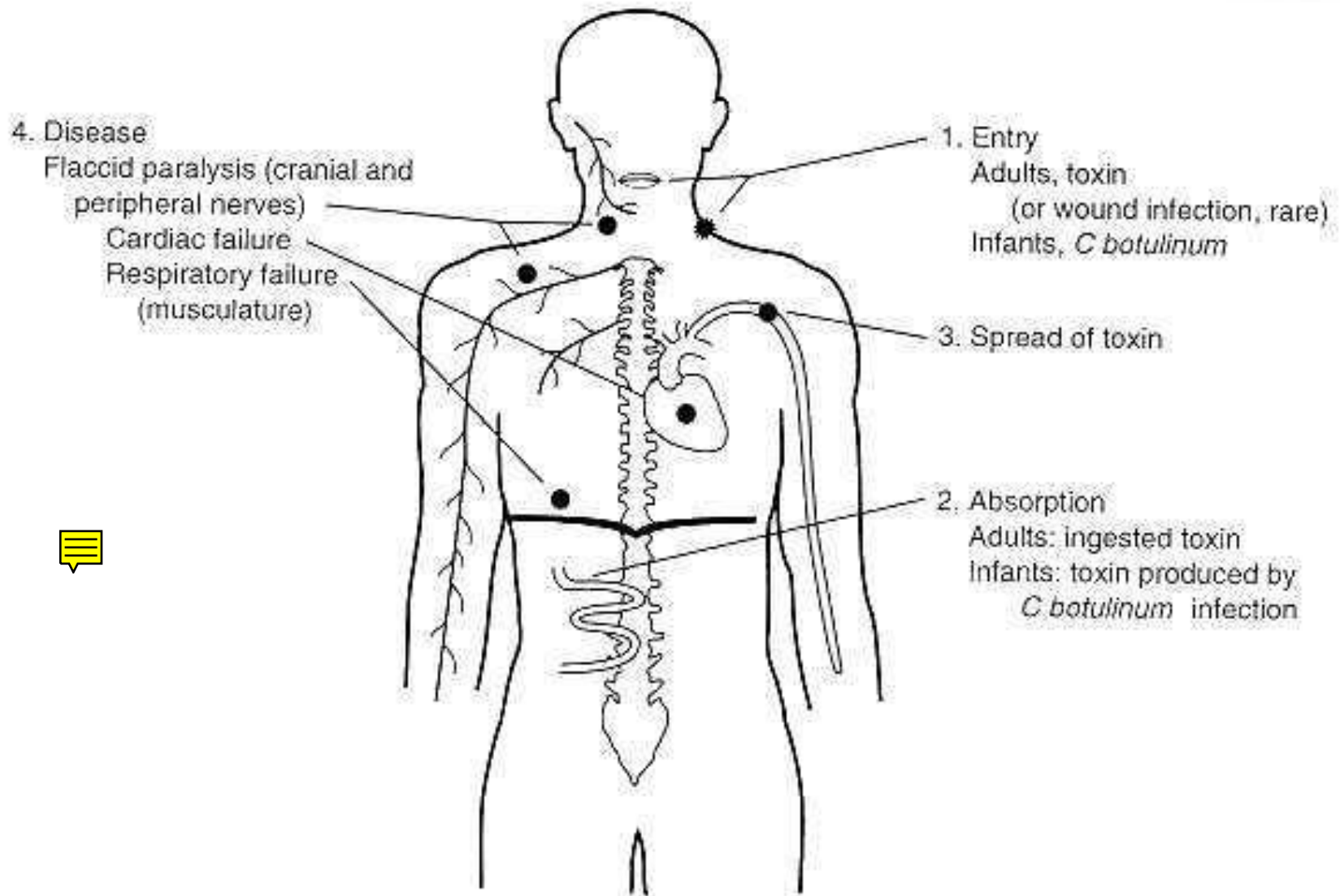
Trouble swallowing, muscle weakness

Due to the flaccid paralysis
Called limp baby syndrome sometimes



Do not feed honey to kids under 2

Clinical Manifestations of *Clostridium botulinum*



Clostridium tetani



- Tetanus
- Rusty nail contaminated with spores punctures skin; wound provides anaerobic environment

STEPPING ON A RUSTED NAIL DOES NOT GIVE YOU TETANIS:

Unless there's spores there

Rusty nail gives you septicaemia

Clean nails can give you tetanus too

- Exotoxin: tetanospasmin



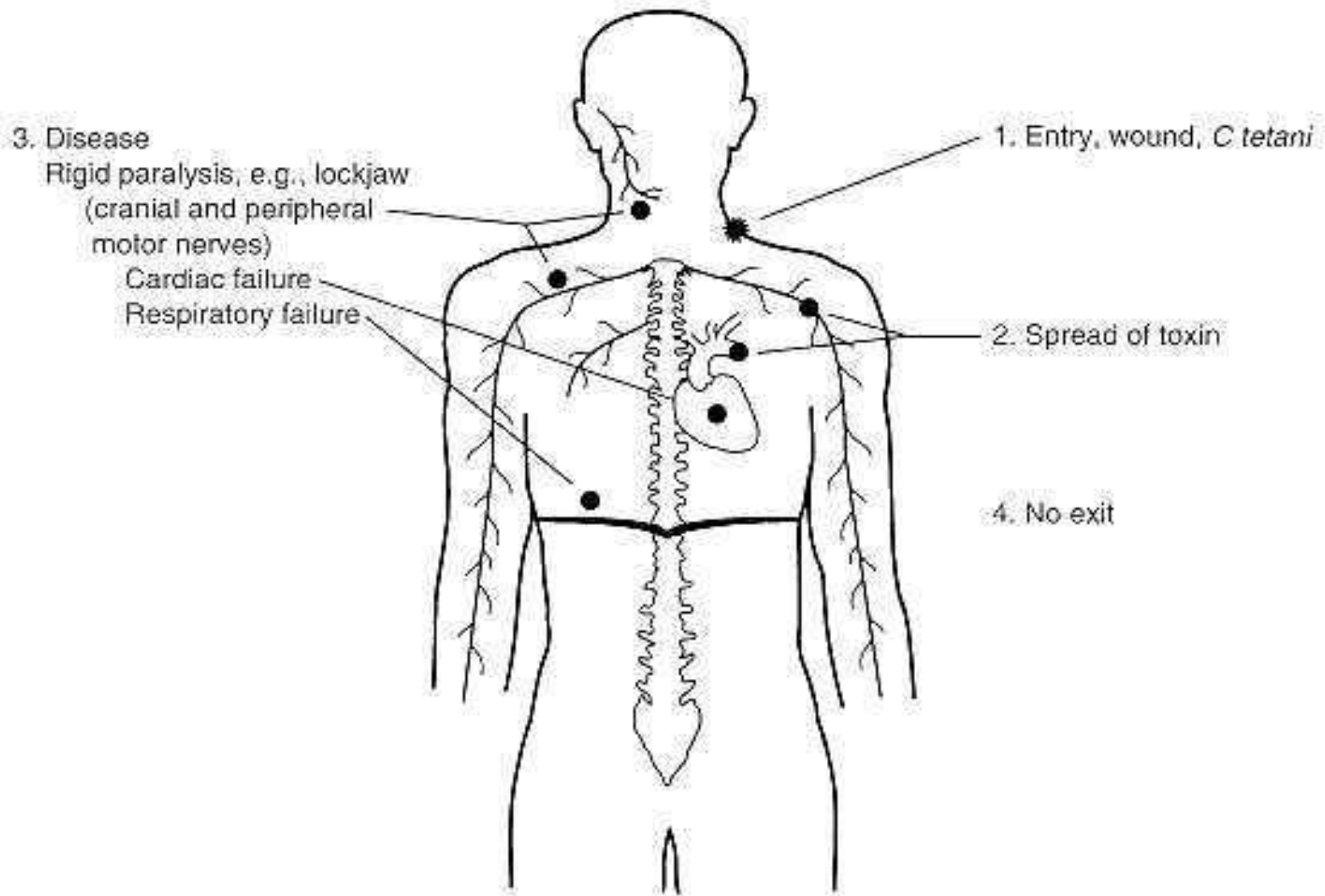
- Sustained contraction of skeletal muscles
- Severe muscle spasms (lock jaw); high mortality at this stage



- Booster (inactivated toxoid) given every 10 years



Clinical manifestations of *Clostridium tetani*



Clostridium perfringens



➤ Gas gangrene

➤ Seen in soldiers wounded in battle

They don't change their socks and boots often which allows for infection

➤ 2 classes of infection:

(i) Wound infection/cellulitis

☰ -necrotic skin exposed to bacteria, damage to local tissues; skin feels moist, spongy, with 'crackly' pockets

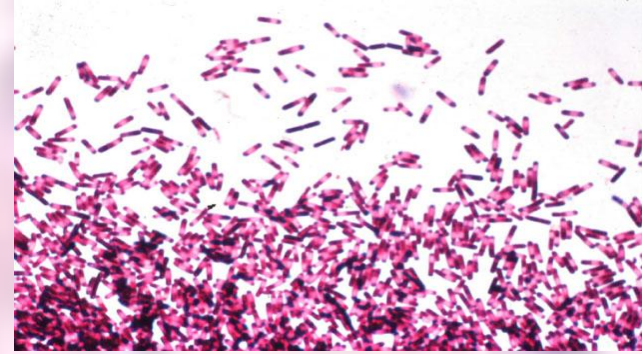
(ii) Clostridial myonecrosis

-bacteria inoculated from trauma into muscles; exotoxin secretion destroys adjacent muscles; as muscles degrade get black fluid excreted from skin

☰ -FATAL unless treated with oxygen, antibiotics (penicillin) and removal of damaged tissue



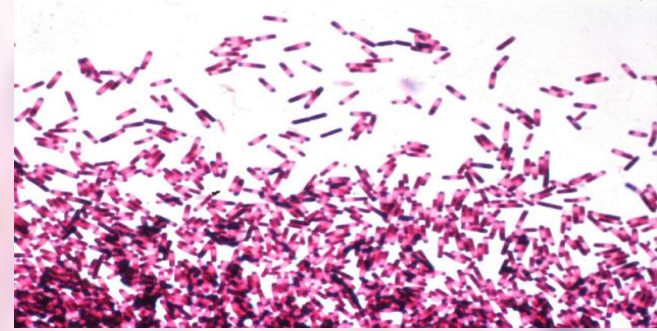
Clostridium difficile



- Causes antibiotic-associated pseudomembranous colitis
- Seen more commonly in hospitals than tetanus, anthrax or botulism
- Overuse of broad-spectrum antibiotics destroys normal intestinal flora Leaving space for bad bacteria
- Infects colon and releases exotoxins
 - Toxin A: diarrhea
 - Toxin B: cytotoxic to colon cells



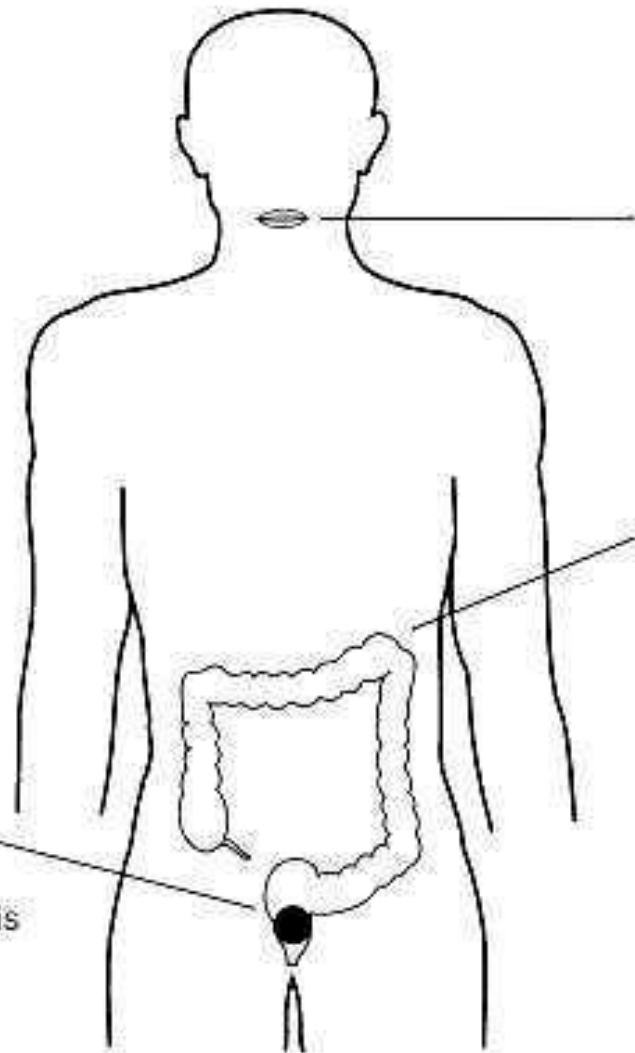
Clostridium difficile



- Symptoms: severe diarrhea, abdominal cramping, fever
- Possible cause of diarrhea in patients on antibiotics
- Treatment
 - discontinue antibiotic treatment
 - Administer metronidazole or vancomycin
 - ?? What is different about these antibiotics ??
You take them orally and they tend to hang out in your gut

Clinical manifestations of *Clostridium difficile*

Studies that indicate that you should be taking a live probiotic to help protect against *C. difficile*



1. Entry

2. Spread

C. difficile overgrowth and toxin production after administration of broad spectrum antibiotics

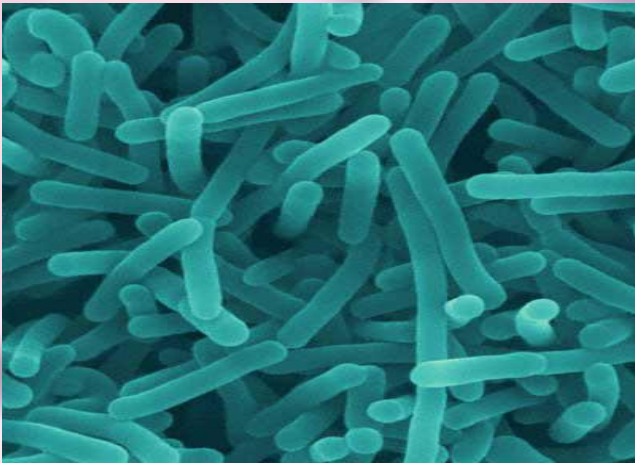
3. Disease

Diarrhea

Pseudomembranous colitis

Non-Spore Forming Rods

- 2 medically important bacilli
 - *Listeria monocytogenes* and *Corynebacterium diphtheriae*

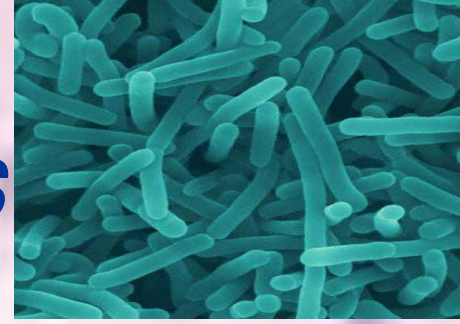


L. monocytogenes



C. diphtheriae

Listeria monocytogenes



- Causative agent of listeriosis; immunocompromised are at high risk
- Found in foods such as soft cheeses, unpasteurized milk, cold cuts, pâté
- **PSYCHROPHILE** → survives in refrigerator
- Variety of symptoms: (depending on how deep it gets inside you)
 - General malaise, diarrhea, meningitis, septicaemia, still-birth/abortions
- Facultative intracellular aerobe
- Crosses 3 protective barriers (blood-brain, GI and feto-placental)
- Treatment: ampicillin or trimethoprim-sulfamethoxazole

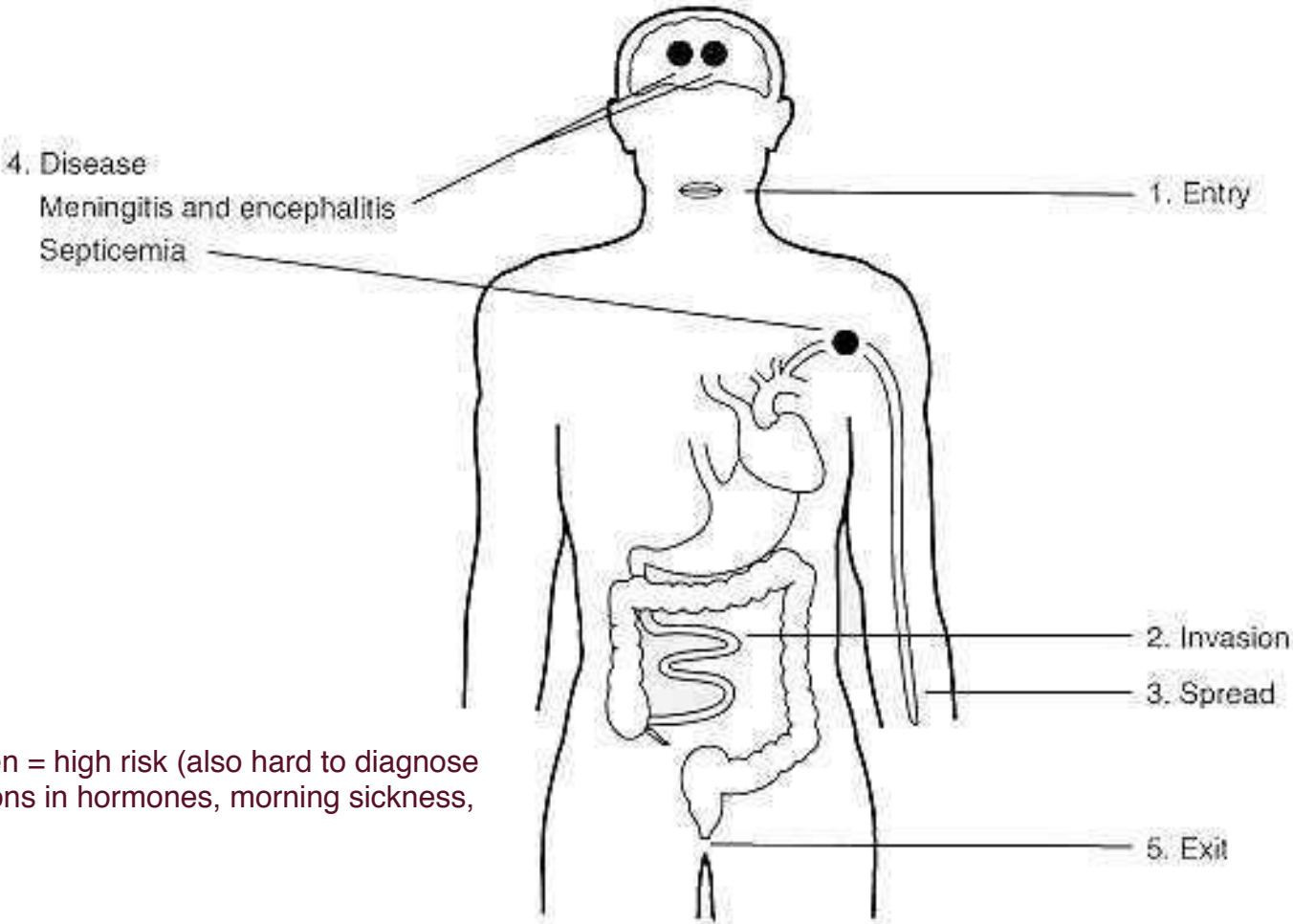


Has not developed resistance to antibiotics

Listeria does not make an exotoxin

Look at the establishment on the package when an outbreak occurs

Clinical manifestations of *Listeria monocytogenes*

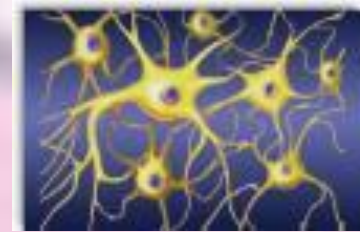
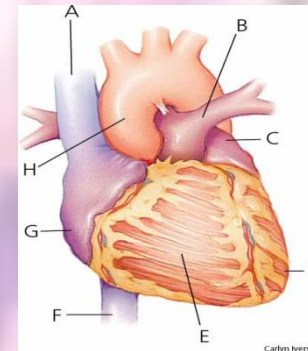


Pregnant women = high risk (also hard to diagnose due to fluctuations in hormones, morning sickness, etc.)

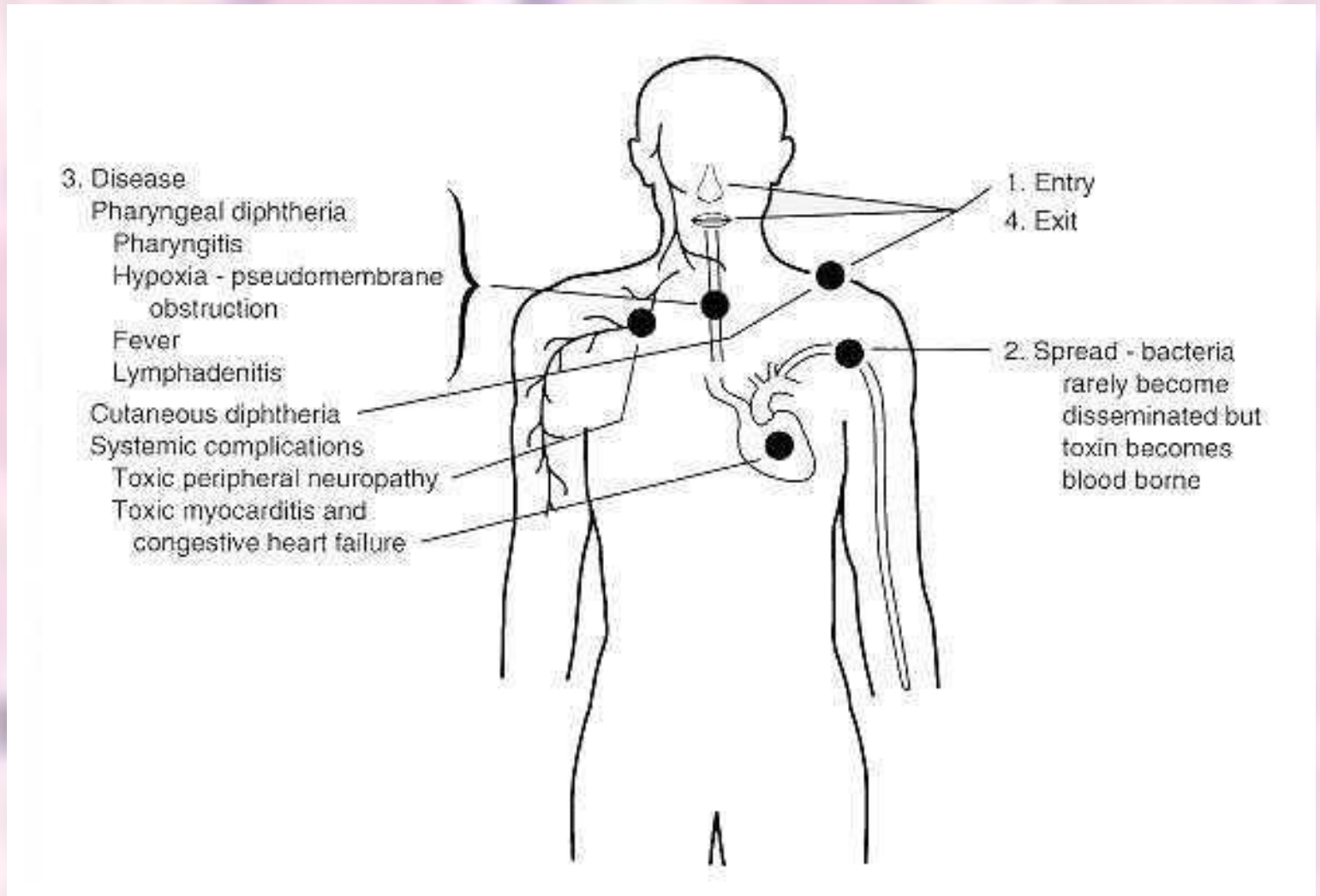
Corynebacterium diphtheriae



- Causative agent of diphtheria
 - Colonization of pharynx and release of exotoxins into bloodstream
- Exotoxin damages heart and neural cells
- Treatment (3 steps):
 - (i) Antitoxin
 - (ii) Penicillin or erythromycin
 - (iii) DPT vaccine
- Can be lysogenized by a bacteriophage (virus that infects bacteria) And then it becomes problematic

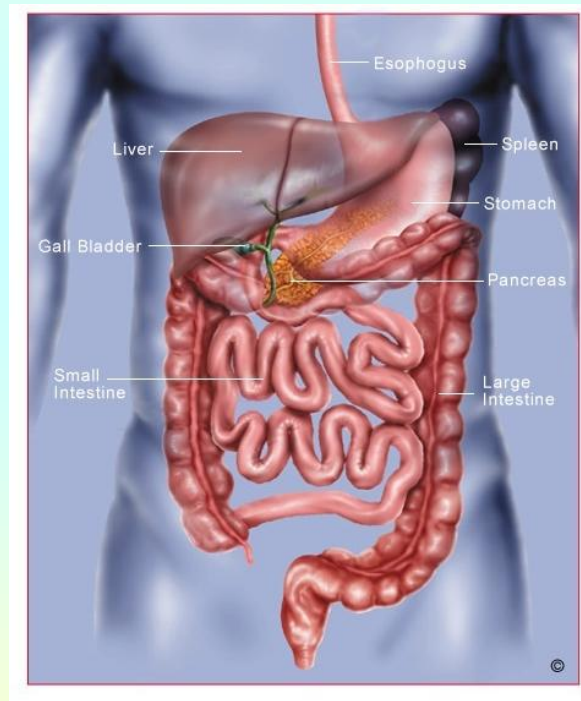


Clinical manifestations of *C. diphtheriae*



Gram Negative Bacilli

The Enterics



Considered enteric because they cause problems in the GI tract

Remember specific, unique things to do with each illness

Enterics

- Found as part of normal intestinal flora BUT can also cause disease
- 4 Major Groups: Enterobacteriaceae (Salmonellae, Shigellae, *E. coli*), Vibrionaceae (*Vibrio*, *Campylobacter*) Pseudomonadaceae (*Pseudomonas*), Bacteroidaceae
- Organisms are divided into groups based on biochemical and antigenic properties

Biochemical Classifications



- Ability to ferment lactose
 - EMB Media:
 - Lactose fermenters are dark purple/black
 - Inhibits Gram positive bacteria
 - MacConkey Media:
 - Lactose fermenters are pink-purple
 - Inhibits Gram positive bacteria



?? What type of media are these ??

SELECTIVE/DIFFERENTIAL

Selective because they have stuff in there that inhibit gram positive

Differential because they can tell whether the colony in there can ferment or something else lactose

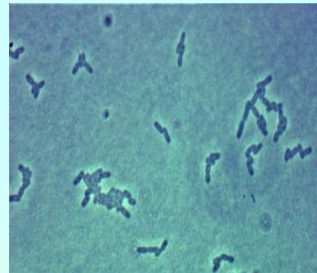
Biochemical Classifications cont'd

DO NOT NEED TO KNOW ANY OF THIS FOR THE EXAM



- H₂S production
- Hydrolysis of urea

- Liquefy gelatin



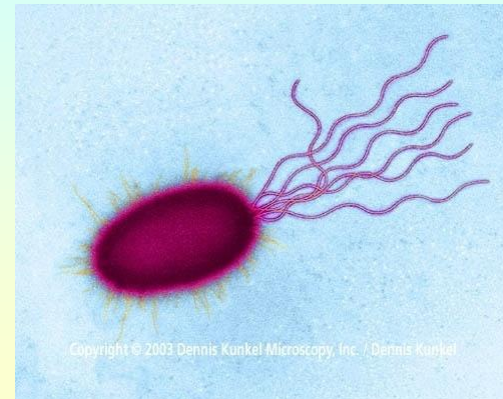
- Decarboxylation of amino acids



Classification Using Surface Antigens

Should know this.

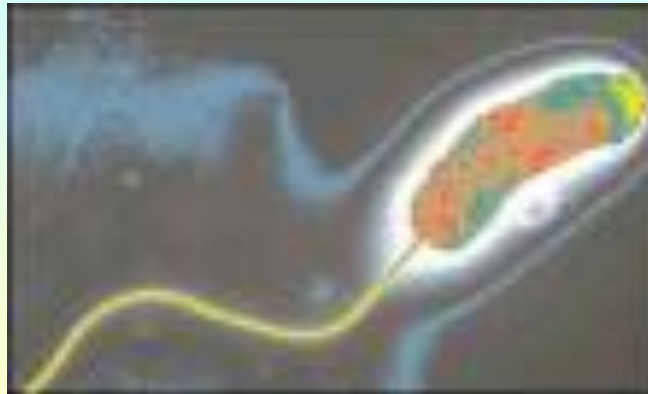
- Variable O-antigen: outermost layer of LPS
 - Changes between enterics
- K-antigen: covers the O-antigen
 - Don't get confused and put C-antigen on the exam!! Its a K for some reason
- H-antigen: flagellar sub-unit
 - Only in motile bacteria



Diseases Caused by Enterics

The deeper they get inside us, the more serious of a disease that can be caused

- Cause diarrhoea with various complications and other infections
- 1) Diarrhea-with/without systemic invasion
 - Bacteria bind intestinal cells but do not enter
 - EXOTOXIN release causes diarrhoea; ENTEROTOXIN causes fluid/electrolyte loss
 - Watery diarrhoea, NO FEVER (because the organism stays attached to the GI tract cell but does not actually get inside)
 - *Vibrio cholera*

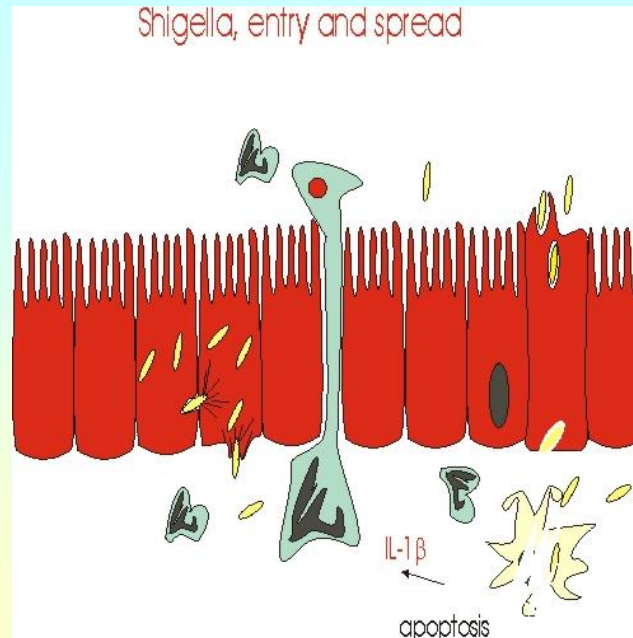


Vibrio cholera

Enteric Diseases cont'd

2) Diarrhea with intestinal cell invasion

- Bacterial virulence factors allow binding and invasion of cells
- Toxin release destroys cells → bloody stools
- Fever response Attach to the GI tract cell and then begin to get in the cell
- *Shigella*,



Enteric Diseases cont'd

3) Diarrhea with invasion of lymph nodes and bloodstream

- Abdominal pain with diarrhoea containing white and red cells
- Fever, headache, increased white cell counts
- *Salmonella Typhi*, *Yersinia enterocolitica*, *Campylobacter jejuni*

Further ability to become blood borne and get to the lymph system



S. Typhi



Y. enterocolitica



C. jejuni

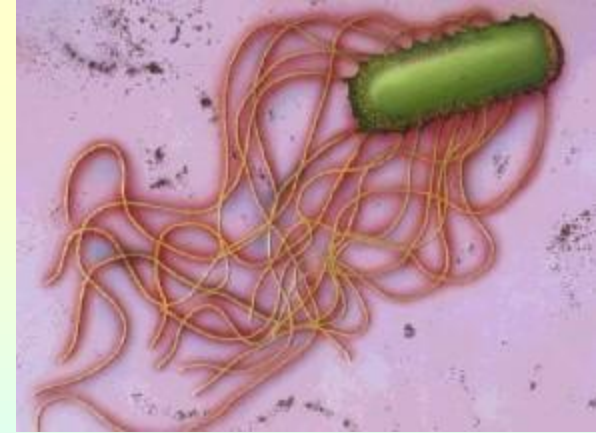
Other Enteric Infections

Some enteric infections do not actually cause diseases in the GI tract:

- Urinary tract infections, pneumonia, bacteremia and sepsis
- Nosocomial infections by: *E. coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Enterobacter*, *Serratia*, *Pseudomonas aeruginosa*
- *Pseudomonas aeruginosa*: opportunistic pathogen, often infects burn patients and can disseminate through body into CNS



Salmonellae



- Member of Enterobacteriaceae family
- Unable to ferment lactose
Appears white - colourless on plate thing
- All have animal reservoirs EXCEPT *S. enterica* serovar Typhi (humans are only host!!)
- Types of infections in humans: enterocolitis, enteric fever, opportunistic infections, septicemia and osteomyelitis

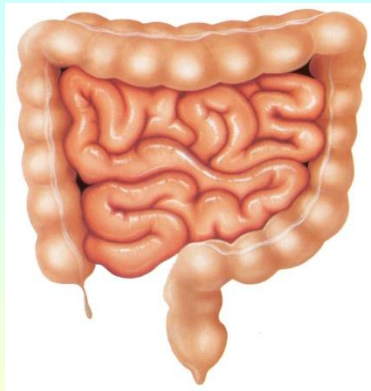
- Two species: *S. enterica* and *S. bongori*
- Enterocolitis (tummy problems):
 - *S. enterica* serovar Enteritidis
 - *S. enterica* serovar Typhimurium
- Enteric fever:
 - *S. enterica* serovar Typhi
 - *S. enterica* serovar Paratyphi

S. enterica serovar Enteritidis

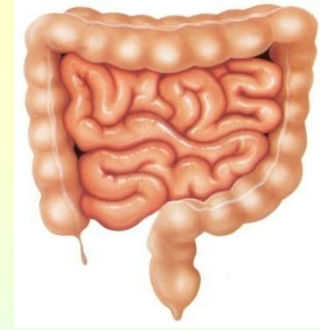
→ Enterocolitis

- **Pathogenesis depends on:**
 - Dose of ingested organism (min 10^5)
 - Immune status of host
 - Virulence of strain
- Incubation time: 6-48h; multiplication in small intestine

Need to ingest a lot



Enterocolitis



- **Symptoms**

- Nausea, vomiting, profuse diarrhoea, abdominal pain
- Fever, chills, headache, myalgia
- 2-3 days recovery
- Septicaemia → Rare



- Use stool culture for lab diagnosis

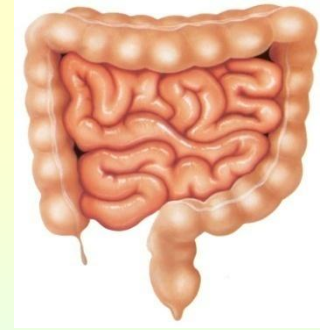
Does not ferment lactose



***Salmonella* on MacConkey**

S. enterica serovar Enteritidis

→ Enterocolitis



- Ingestion of contaminated food
- Poultry, eggs, meat and milk



- Person to person spread
- Most cases occur at home
- Under-reported and undiagnosed
- Antibiotics **NOT RECOMMENDED** → WHY?

Because salmonella actually grows better with the antibiotic

Enteric Fever



- *S. enterica* serovar Typhi → typhoid fever
- *S. enterica* serotypes Paratyphi A, Schottmuelleri, Hirschfeldii → paratyphoid fever (milder)
- **Enteric fever**: generalized infection; bacterial multiplication in lymphoid tissue
- Necrosis of intestinal lymphoid tissue ^{Death} → ulceration, haemorrhage, perforation
- **Untreated**: 10% mortality won't ask about statistics
- **Convalescent carriers**: excrete bacteria for 3 months
- **Chronic carriers** (1-2%): excrete bacteria for at least 6 months, sometimes life long



Enteric Fever

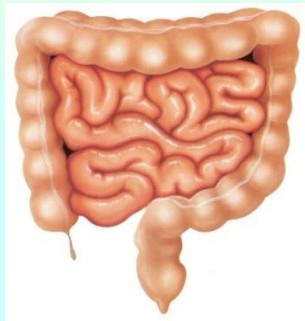


- Diagnosis: Isolation of bacteria from blood (1st week)
Stool and urine (2nd-3rd week)
- Infective dose is 10^6 organisms
- Sources are contaminated drinking water, shellfish, milk and milk products
- Clean handling of food, water treatment and safe sewage disposal are essential
Know the difference between salmonella that causes tummy problems and the blood one and typhie and paratyphi
- Vaccine available, but only effective against small bacterial load
Antibodies can't get rid of all of them

Escherichia coli



- Most numerous aerobic bacteria of normal gut flora




- Lactose fermenting



***E. Coli* on
MacConkey agar**

- Pathogenic to other parts of the body
 - responsible for 85% of bacteriuria
Bacteriuria = bacteria in the urine

E. coli → Gastroenteritis

- Enterotoxigenic *E. coli* Makes and enterotoxin
 - Infant diarrhoea (developing countries)
 - Traveller's diarrhoea 
 - Enterotoxins



- Enteroinvasive *E. coli*
 - Symptoms similar to shigellosis

E. coli → Gastroenteritis

- Enteropathogenic *E. coli*
 - older name for some serotypes causing infant diarrhoea
- *E. coli* O157:H7 Remember this one
 - Haemorrhagic colitis
 - Hamburger disease
 - Proper handling of food, safe preparation and proper cooking practices are essential to prevent illness Can cause HUS (haemolytic urigmiate syndrome?) which causes kidney failure, usually in kids
- *E. coli* is also implicated in neonatal meningitis and nosocomial urinary and wound infections



O means...
H7 has to do with flagellum

This strain is a problem because every cow has it (it does not affect cows)
The challenge is removing this bacteria (many people have tried it) (Feeding the cow a specific bacteriophage a couple days before slaughter helps get rid of it, paired with hot wash after slaughter)

Shigellae



- Cause acute diarrhoea with mucus, pus and blood
- Generally non-lactose fermentors
- Dose of 10^5 infects 25% of people, 10^9 infects 95%
- *Shigella sonnei* → Europe and North America

- *S. dysenteriae*:



- Tropics
 - **SEVERE** illness: watery diarrhoea, cramps, fever
 - Infection from **SMALL** numbers of organism
- Most commonly seen in children; poor sanitation and crowding
 - Prevention by safe handling of food, treatment of water, safe disposal of sewage
 - **NO VACCINE**





Vibrio cholerae



- Causes cholera: acute gastrointestinal illness
- Profuse watery diarrhoea, cramps and vomiting
- Enterotoxin binds cells in small intestine
 - Cells secrete chlorides, ↓ Na⁺ absorption
 - Water accumulates in gut → watery diarrhoea
 - Can lead to **severe** dehydration and death if untreated
- Endemic in South East Asia and parts of Africa
 - Lack of clean drinking water!
- Mainly water-borne MASSIVE ELECTROLYTE IMBALANCE
- Massive (10-15 litres per day) loss

Big thing to remember → need to replenish water drastically or else death can occur



Campylobacter



- *C. jejuni* and *C. coli*
- Major cause of human enteritis
- Normal flora in birds and domestic animals
- Some strains invasive, others toxigenic
- Symptoms: fever, abdominal pain, bloody diarrhoea
- Maybe one cause of traveller's diarrhoea



For this class, traveller's diarrhoea is related to E.coli

Microaerophilic (makes it hard to isolate and grow in the lab)

Pseudomonas



- Opportunistic pathogen
- Found in many moist habitats and water
- Source of infection can be humidifiers etc.
- Treatment is difficult because all *Pseudomonas* *sp.* are resistant to many antibiotics!!!!

inactivating enzymes

Pseudomonas aeruginosa

Categorized as an enteric but it can cause problems in the lungs

- Respiratory pathogen in cystic fibrosis patients



- Infections in lesions of burn patients

Burn patients can become easily infected



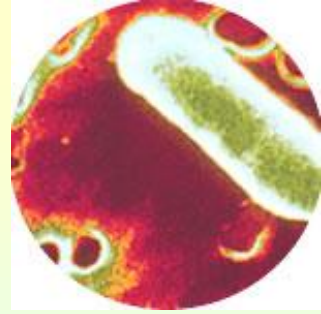
Pseudomonas cepacia

- Common contaminant of saline solutions and water
- Able to multiply in low nutrient environment
- Respiratory pathogen of cystic fibrosis patients



Remember the two Pseudomonas
Grape-like odour signals a Pseudomonas infection

Haemophilus influenzae



- Part of normal nasopharyngeal flora in many adults and children
- Causes invasive infections of young children
 - Meningitis, pneumonia, joint infections
 - Development of vaccine, now used routinely, decreased the number of cases in Canada
- Can cause increased bronchial inflammation in patients already having chronic bronchitis



Enterobacter spp.

- Can cause nosocomial infections



- Wound infections, pneumonia, bacteremia
- *E. sakazakii* linked to infant illness from powdered infant formula

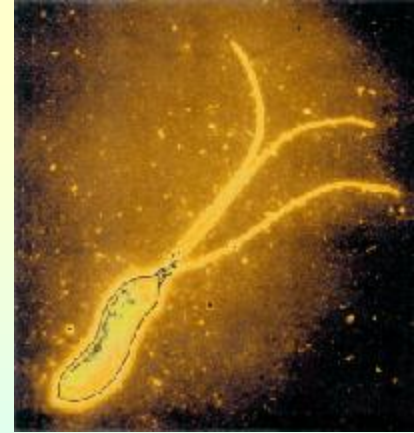


Powdered infant formula is not a sterile product

Low birth-weight babies are a big problem with this

Now called *Cronobacter*

Helicobacter pylori



- Microaerophilic, spiral bacilli
- Most common cause of stomach ulcers
 - In the past, cause was thought to be stress and diet
 - 1982-Dr. Robin Warren and Dr. Barry Marshall discovered link between *H. pylori* and ulcers
 - Medical community slow to accept their theory (1994- National Institute of Health Conference concludes strong association between ulcers and *H. pylori*)
- Urease: protection from low pH
- Triple therapy treatment: antibiotics and H⁺ pump inhibitors



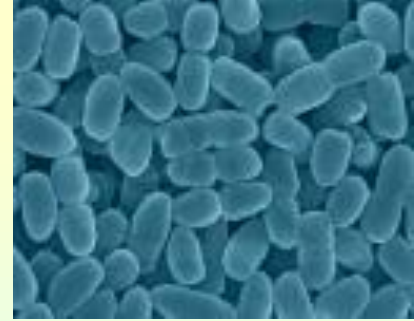
Can be detected using breath test

Hard to prove that this was actually the cause of ulcers because the stomach is so acidic that the bacteria shouldn't be able to survive, but it does

Duodenal Ulcer

Bordetella pertussis

Also causes respiratory problems



- Whooping cough ***VIOLENT COUGH***
- **4 Virulence Factors** Do not need to remember what the toxins are, just remember that the bacteria makes a lot of toxins and what they do
 - Pertussis toxin (A-B)
 - Extra cytoplasmic adenylate cyclase (weakens host defense)
 - Filamentous hemagglutinin (bronchial attachment and exotoxin release)
 - Tracheal cytotoxin (destroys ciliated cells → poor clearance of mucous and bacteria)
- Prevention: vaccination with heat-killed organism





Legionella pneumophila



- Causes Legionnaires disease 🗨
- Opportunistic pathogen
- May cause severe pneumonia
- Grows in water and is found in shower heads, water tanks, air cooling/heating tanks - this is how it becomes aerosolized
- Exposure is by aerosol and there is **NO** person-to-person transmission

Cannot catch it by someone coughing on you